

Claims

1. *Xenorhabdus bovienii* strain H31 deposited with NCIMB under accession number NCIMB 40985.

2. *Xenorhabdus bovienii* strain I73 deposited with NCIMB under accession number NCIMB 40986.

3. A pesticidal agent which (i) is obtainable from a *X. bovienii* strain; (ii) has oral insecticidal activity against one or more species of insect of the order Lepidoptera, Coleoptera or Homoptera; (iii) is substantially heat stable to 50°C; and (iv) acts synergistically with *B. thuringiensis* cells as an oral insecticide.

4. An agent as claimed in claim 3 wherein the agent has activity against insects of two or more orders selected from: Lepidoptera; Coleoptera; Homoptera.

5. An agent as claimed in claim 3 or claim 4 which is obtainable from a strain as claimed in claim 1 or claim 2.

6. An agent as claimed in claim 5 which is H toxin or I toxin

7. An isolated nucleic acid comprising a nucleotide sequence encoding an agent as claimed in claim 5 or claim 6.

8. A nucleic acid as claimed in claim 7 comprising a nucleotide sequence identical to any one or more of I73APT.seq, I73BPT.seq, I73CPT.seq, and I73DAPT.seq or a sequence degeneratively equivalent thereto.

9. An isolated nucleic acid comprising a homologous variant of the nucleotide sequence of claim 7 or claim 8 having about 70% or more sequence identity therewith and encoding a polypeptide which is an insecticidal toxin.

10. A nucleic acid as claimed in claim 9 wherein the variant is an insecticidal toxin obtainable from a bacterial nematode-symbiont.

11. A nucleic acid as claimed in claim 9 having a sequence which is a derivative of the nucleotide sequence of claim 7 or claim 8 by way of addition, insertion, deletion or substitution of one or more nucleotides.

12. A nucleic acid as claimed in any one of claims 7 to 11 further encoding a pesticidal material derived from *Bacillus thuringiensis*.

13. A nucleic acid as claimed in claim 12 wherein the agent and the pesticidal material derived from *Bacillus thuringiensis* are expressed as a fusion protein

14. A nucleic acid which is complementary to the nucleic acid of any one of claims 7 to 13.

15. A nucleic acid molecule for use as a probe or primer, said molecule having a nucleotide sequence of at least 15, 18, 21, 24 or 30 nucleotides, which sequence is present in, or complementary to, the nucleic acid of claim 7 or claim 8.

16. A method for identifying or cloning an insecticidal toxin, which method employs a nucleic acid molecule having a nucleotide sequence as claimed in claim 15.

17. A method as claimed in claim 16 comprising the steps of:  
(a) providing a preparation of nucleic acid from a bacterium,  
(b) providing a probe as claimed in claim 15,  
(c) contacting nucleic acid in said preparation with said probe under conditions for hybridisation of probe to any said gene or homologue in said preparation, and,  
(d) identifying said gene or homologue if present by its hybridisation with said probe.

18. A method as claimed in claim 17 wherein the hybridisation conditions are selected to allow the identification of sequences having about 70% or more sequence identity with the probe.

19. A method as claimed in claim 16 comprising use of two primers to amplify a nucleic acid encoding an insecticidal toxin.

20. A method as claimed in claim 16 or claim 19 comprising the steps of :

- (a) providing a preparation of nucleic acid from a bacterium,
- (b) providing a pair of nucleic acid molecule primers, at least one of which is a primer as claimed in claim 15,
- (c) contacting nucleic acid in said preparation with said primers under conditions for performance of PCR,
- (d) performing PCR and determining the presence or absence of an amplified PCR product.

21. A method as claimed in any one of claims 17 to 20 wherein the bacterium is obtained from a nematode identified as being pathogenic to an insect.

22. A recombinant vector comprising the nucleic acid of any one of claims 7 to 13.

23. A vector as claimed in claim 22 which is capable of replicating in a suitable host.

24. A vector as claimed in claim 23 which is a baculovirus.

25. A vector as claimed in claim 23 wherein the nucleic acid is operably linked to a promoter or other regulatory element for transcription in a host cell.

26. A vector as claimed in claim 25 further comprising any one or more of the following: a terminator sequence; a polyadenylation sequence; an enhancer sequence; a marker gene; a sequence encoding pesticidal material derived from *Bacillus thuringiensis*.

27. A vector as claimed in claim 25 or claim 26 which is a plant vector.

28. A method comprising the step of introducing a vector as claimed in any one of claims 23 to 27 into a cell.

29. A method for transforming a plant cell, comprising a method as claimed in claim 28, and further comprising the step of causing or allowing recombination between the vector and the plant cell genome to introduce the nucleic acid into the genome.

30. A host cell comprising a vector as claimed in any one of claims 25 to 27.

31. A host cell transformed with a vector as claimed in any one of claims 25 to 27.

32. A host cell as claimed in claim 30 or claim 31 which is a plant cell.

33. A host cell as claimed in claim 32 which is in a plant.

34. A method for producing a transgenic plant comprising a method as claimed in claim 28 and further comprising the step of regenerating a plant from the transformed cell.

35. A plant comprising the cell of claim 32 to claim 33.

36. A plant as claimed in claim 35 produced by the method of claim 34.

37. A plant which is the progeny of a plant as claimed in claim 35 or claim 36.

38. A plant as claimed in any one of claims 35 to 37 which is selected from: maize, cotton, soya, rice, tomato, potato and *Brassica* species.

39. A part or propagule of the plant of any one of claims 35 to 38.

40. A polypeptide encoded by the nucleic acid of any one of claims

7 to 13.

41. A method of producing a polypeptide comprising the step of causing or allowing the expression from a nucleic acid of any one of claims 7 to 13 in a suitable host cell.

42. A method of influencing or affecting the toxicity of a plant cell, the method including causing or allowing expression of a heterologous nucleic acid as claimed in any one of claims 7 to 13 within the cells of the plant.

43. A composition comprising the polypeptide of claim 40.

44. A composition as claimed in claim 43 which is adapted for oral administration.

45. A composition as claimed in claim 44 further comprising pesticidal material derived from *Bacillus thuringiensis*

46. Use of a material selected from: an *X. bovienii* strain of claim 1 or claim 2; an agent of any one of claims 3 to 6; a nucleic acid of any one of claims 7 to 13; a vector of any one of claims 23 to 27; a host cell of any one of claims 30 to 33; a plant of any one of claims 35 to 38; a polypeptide of claim 40; or a composition of any one of claims 43 to 45; for the control of a pest.

47. Use as claimed in claim 46 wherein the pest is an insect and the material is used to kill the insect.

48. Use as claimed in claim 46 or claim 47 wherein the pest is selected from: *Leptinotarsa decimlineata*; *Diabrotica undecimpunctata*; *Diabrotica virgifera*; *Anthrenus grandis*; *Plutella xylostella*; *Heliothis virescens*; *Pieris rapae*; *Ostrinia nubilalis*; *Spodoptera exigua*.

49. Use of an agent of any one of claims 3 to 6; a host cell of any one of claims 30 to 33; a plant of any one of claims 35 to 38; a polypeptide of claim 40; or a composition of any one of claims 43 to 45; optionally in conjunction with *Bacillus thuringiensis* or

pesticidal materials derived therefrom as an oral pesticide.

50. An antibody or fragment thereof, or a polypeptide comprising the antigen-binding domain of the antibody, capable of specifically binding the polypeptide of claim 40.

51. A method of producing the antibody or fragment as claimed in claim 50 comprising the step of immunising a mammal with a polypeptide according to claim 40.

52. A method of identifying and/or isolating an insecticidal toxin comprising the step of screening candidate polypeptides with a polypeptide comprising the antigen-binding domain of the antibody of claim 50.

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